Data Evaluation Record on the Fish Short-Term Reproduction Assay with Cypermethrin

EPA MRID Number 48683001

Data Requirement:	EPA DP Barcode	401651
	OECD Data Point	229
	EPA MRID	48683001
	EPA Guideline	890.1350, Fish Short-Term Reproduction Assay

Test material: Cypermethrin **Purity:** 95.2%

Common name

Chemical name: IUPAC: mixture of the stereoisomers (S)-α-cyano-3-phenoxybenzyl (1RS,3RS;1RS,3SR)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate where the ratio of the (S);(1RS,3RS) isomeric pair to the (S);(1RS,3SR) isomeric pair lies in the ratio range 45-55 to 55-45 respectively

CAS name: (S)-cyano(3-phenoxyphenyl)methyl 3-(2,2-dichloroethenyl)-2,2-dimethylcyclopropanecarboxylate.

CAS No.: 52315-07-8

Synonyms: FMC 30980

EPA PC Code: 109703

Primary Reviewer: Joan Gaidos — Signature: — Date: 1/07/13

Senior Scientist, Cambridge Environmental, Inc.

Secondary Reviewer: Teri S. Myers — Signature: — Date: 2/25/13

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EPA/EFED/ERB6

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Additional Reviewer(s): Amy Blankinship, Chemist
EPA/EFED/ERB6

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Date Evaluation Completed: 6/6/15

CITATION: York, D.O. 2012. Cypermethrin – Fish Short-term Reproduction Assay with the Fathead Minnow (*Pimephales promelas*). Performed by Smithers Viscient, Wareham, Massachusetts, Laboratory Project No. 1781.6766. Submitted by Syngenta Ltd, Jealott's Hill International Research Centre, Bracknell, Berkshire RG42 6EY, United Kingdom; FMC Corporation, Philadelphia, Pennsylvania; United Phosphorus, Inc., King of Prussia, Pennsylvania. Completion date April 18, 2012.

Note: The US EPA Endocrine Disruptor Screening Program (EDSP) Tier 1 screening battery is comprised of eleven screening assays intended to identify a chemical's likely endocrine bioactivity, i.e., its potential to interact with the estrogen, androgen, or thyroid (E, A, or T) pathways. The robustness of the Tier 1 battery is based on the strengths of each individual assay to identify potential endocrine bioactivity with complementary endpoints within the assay, where available, and redundancy across the battery. Thus, the results of each individual assay should not be considered in isolation but rather should be considered in the context of other assays in the battery as well as Other Scientifically Relevant Information (OSRI). In order to determine if e chemical has the potential to interact with the E, A or T pathweys, a Weight of Evidence (WoE) evaluation of Tier 1 assay results, in combination with the findings in the OSRI, should be undertaken (refer to the WoE Document).

Guideline recommendations are provided in italics; these recommendations should remain visible in the completed DER.

Disclaimer: The guideline recommendations in this DER template are offered as a general reference to aid in preparation of the DER. The purpose of these recommendations is not to serve as substitute for the Test Guidelines, nor to provide any guidance on how the study should be conducted.

EXECUTIVE SUMMARY:

The 21-day short-term reproduction assay of cypermethrin with fathead minnows (*Pimephales promelas)* was conducted under flow-through conditions. Adult fish (16 spawning groups, 2 males and 4 females in each group; 5 months old) were exposed to cypermethrin (95.2% purity) at nominal concentrations of 0 (control), 0.000030, 0.00030, and 0.003 mg a.i./L; mean-measured concentrations were <LOQ (<0.000001), 0.000013, 0.00012, and 0.0014 mg a.i./L. The test system was maintained at 25 to 26°C and a pH of 7.0 to 7.7.

There were no significant differences in either male or female survival at any treatment levels compared to the negative control. Percent survival was 100% for all groups of males and was 100, 94, 94, and 88% for the negative control and low, mid, and high treatment concentrations, respectively, for females. During the in-life exposure, there were no notable observations that occurred with regards to behavior, coloration/banding, changes in ovipositor appearance, or size of the dorsal nape pad, and no clinical signs of toxicity were observed. There were no treatment-related effects on male and female body weight and length.

Spawning occurred at least every 4 days in thee of the four control replicates and mean fecundity in control averaged 14 eggs/female/day/replicate (range: 13-18 eggs/female/day among the replicates); fertilization success in the control group averaged 96%. Fecundity was decreased ($p<0.05$) by 42% and 84% in the mid- and high-treatment females, respectively. There were no treatment-related effects ($p>0.05$) on fertilization success.

Male gonadal somatic index (GSI) was increased ($p<0.05$) by 22 and 27% at the mid and high treatment concentrations, respectively, compared to the negative control. There were no treatment-related effects on female GSI ($p>0.05$) or male and female plasma vitellogenin (VTG). Additionally, there were no significant effects on median male tubercle score, and no tubercles were observed on any females.

Although not analyzed statistically, in male fish, there were only background or sporadic gonadal histopathological findings noted that were not considered related to treatment. In female fish, there was an increased incidence of mature oocyte atresia at the high treatment concentration, which was associated with

granulomatous inflammation and the presence of microsporidia in half the observed cases across all treatment levels. There were no significant findings observed in the gonadal stage for either males or females, and no notable observations in secondary sex characteristics for any treatment group compared to the controls.

The performance criteria for fecundity was not met. There were one replicate in the control group (Replicate C) where there was a period of 4 days in which no spawning occurred and fecundity was 13 eggs/female/day. Therefore, this replicate did not meet the criteria of either at least 15 eggs/female/day or spawning at least every 4 days. However, it is noted that the overall control mean fecundity value (14) was just slightly below 15 eggs/female/day, and the other two replicates did achieve 15 or greater eggs/female/day and the remaining replicate spawned at least every 4 days (11 eggs/female/day). Also, the %CV values for the mean recoveries at the two highest treatment concentrations were 45.6 and 51.2%, which did not meet the validity criteria of <20% over the 21-day test. There was no pattern of decline during the study period and no un-dissolved test substance was observed in the dilution system, as such, the recoveries indicate the test material was generally poorly recovered in solution under the test conditions. Given the physicochemical properties of cypermethrin, these results may be considered consistent with expectations for the chemical and previous experience with the test item. The remaining validity criteria for OCSPP 890.1350 were met. Therefore, while these deviations are noted, they did not sufficiently impact the interpretation of the assay results.

There was one fish that was inadvertently counted as a female but later discovered to be a male. This occurred in one of the control replicates and data from that replicate were subsequently reanalyzed for the gender specific endpoints of male GSI, male VTG, body weight, and length. This did not have an impact on the conclusions of the study as male GSI remained the only male gender specific endpoint with significant effects.

This assay satisfies the EDSP Tier 1 Test Order requirements for a Fish Short-Term Reproduction Assay (OCSPP Guideline 890.1350).

Results Synopsis

Test Organism age at test initiation: *ca.* 5 months

Mean body weight at test initiation (if measured): Male 3.2 g; Female 1.7 g

Mean length at test initiation (if measured): Not reported.

Test Type (Flow-through, Static, Static Renewal): Flow-through

Table 1: Summary of Reproductive and HPG Effects[1,2] in the Fish Short-Term Reproduction Assay (FSTRA) with Cypermethrin.

Treatment (mg a.i./L) [mean-measured]	Fecundity	Fert. Success	Tubercle Score		GSI		Gonadal Histo.		Plasma VTG		Plasma T		Plasma E2	
			M	F	M	F	M	F	M	F	M	F	M	F
0.000013	No	No	No	NA	No	No	NA	NA	No	No	NA	NA	NA	NA
0.00012	Yes	No	No	NA	Yes	No	NA	NA	No	No	NA	NA	NA	NA
0.0014	Yes	No	No	NA	Yes	No	NA	NA	No	No	NA	NA	NA	NA

Abbreviations: Conc. Concentration. Diff. Difference. E2 17β-estradiol. F Female. Fert. Fertilization. GSI Gonado-Somatic Index. Histo. Histopathology. M Male. NA Not applicable. T Testosterone. VTG Vitellogenin.

1 A "yes" indicates a significant difference based on comparison to the negative (clean water) control, unless otherwise specified.

2 The criteria for significance are described in the Reviewer's Analysis and Statistical Verification sections of the DER. Conclusions regarding histopathology may be heavily weighted by the expert opinion of a board-certified pathologist.

I. MATERIALS AND METHODS

GUIDELINE FOLLOWED: This study was conducted according to the U.S. EPA OCSPP 890.1350: "Fish Short-Term Reproductive Assay" and OECD 229 (2009). The following deviations were noted:

1. There were one replicate in the control group (Replicate C) where there was a period of 4 days in which no spawning occurred, and fecundity was 13 eggs/female/day, respectively. Therefore, this replicate did not meet the criteria of either at least 15 eggs/female/day or spawning at least every 4 days. However, it is noted that the overall control mean fecundity value (14) was just slightly below 15 eggs/female/day, and the other two replicates did achieve 15 or greater eggs/female/day and the remaining replicate spawned at least every 4 days (11 eggs/female/day).
2. Analytical verification of the test material from Days 0, 6, 11, 19 and 21 yielded recoveries of 44% to 67% of nominal concentrations. Results of these analyses indicate measured concentrations where generally lower than nominal, but according to the study author were consistent with expectations for the physicochemical properties and previous experience with the test item. Therefore, although there was no pattern of decline during the study period and no un-dissolved test substance was observed in the dilution system, the recoveries indicate the test material was generally poorly recovered in solution under the test conditions. The %CV values for the mean recoveries at the two highest treatment concentrations were 45.6 and 51.2%, which did not meet the validity criteria of <20% over the 21-day test. The test material was detected on Days 11 (one replicate) and 19 (two replicates) in the control samples. The study author's concluded the contamination was likely a result of processing samples after being removed from the exposure system and that the control fish were not exposed to cypermethrin.
3. The physico-chemical properties of the test material were not reported.

4. The unionized ammonia and residual chlorine in the test water were not reported. The OCSPP 890.1350 performance criteria establish maximum levels for these values and it is unclear if the maximum recommendations were exceeded.
5. Incorrect sexing of individuals in one replicate from the control group resulted in there being three males and three females in this replicate instead of the two males and four females specified in the OCSPP 890.1350 guideline.

These deviations do not sufficiently impact the acceptability of the study.

COMPLIANCE: Signed and dated No Data Confidentiality, GLP, and Quality Assurance statements were provided. This study was conducted in compliance with all pertinent U.S. EPA Good Laboratory Practice regulations with the following exceptions: routine dilution water and food contaminant screening analyses were conducted following standard validated methods. This exception has no impact on the study results.

A. TEST MATERIAL: Cypermethrin; CAS# 52315-07-8.

Description: Amber liquid.

OECD recommends describing water solubility, melting/boiling point stability in water and light, pKa, Pow or Kow, vapor pressure of test compound, expiration date.

Lot No./Batch No. : PL10-0492

Purity: 95.2%

Impurities: None reported

Stability of Compound: Analytical verification of the test material from Days 0, 6, 11, 19 and 21 yielded recoveries of 44% to 67% of nominal concentrations. Stock solution (FETAX) fortified with cypermethrin at concentrations of 0.000003, 0.00005, and 0.0005 mg a.i./L yielded recoveries of 80.9 to 96.8% of nominal concentrations. Quality control samples (n=17) fortified at 0.000015, 0.0003, and 0.003 mg a.i./L yielded recoveries of 83.0 to 118% of nominal concentrations. Results of these analyses indicate measured concentrations where generally lower than nominal, but according to the study author were consistent with expectations for the physicochemical properties and previous experience with the test item. Therefore, although there was no pattern of decline during the study period and no un-dissolved test substance was observed in the dilution system, the recoveries indicate the test material was generally poorly recovered in solution under the test conditions. Additionally, the %CV values were 45.6 and 51.2% for the 0.00012 and 0.0014 mg a.i./L treatment concentrations, respectively, and did not meet the validation criteria of <20%.

Storage Conditions of Test Chemicals: Stored at room temperature in a dark, ventilated cabinet.

B. Test organism:

Table 2: General Information About the Test Species and Acclimation.

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Species common name:	Fathead Minnow		*EPA recommends fathead minnow (Pimephales promelas).*
Species scientific name:	*Pimephales promelas*		
Species strain (if stated):	Not reported		
Were fish obtained from a single laboratory stock?	Yes		*EPA recommends that fish be from a single laboratory stock.*
Were acclimation conditions same as definitive test?	Yes		*EPA recommends that fish be acclimated under water quality and illumination conditions that are similar to the definitive test.*
Acclimation period:	16 days		*EPA recommends a minimum two-week acclimation period. Note that the acclimation period is different from the subsequent, in situ pre-exposure phase.*

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Details on health:		No mortalities were reported during the 7 days prior to pre-exposure period. Fish did not receive any treatment for disease in the two weeks prior to spawning (pre-exposure period). Behavioral abnormalities or clinical signs were not reported.	*EPA recommends that mortality during the 7 days prior to the pre-exposure phase be less than 5% of the culture population. If mortality during these 7 days is greater than 10%, EPA recommends that the fish be rejected. If mortality is between 5-10%, EPA recommends that fish be held another 7 days. If mortalities greater than 5% occur during this extended acclimation period, EPA recommends that the fish not be used.*

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Type of food:	Frozen brine shrimp		*EPA recommends that fish be fed frozen brine shrimp twice per day to promote active reproduction and maintain body condition.*
Source of food:	Not reported	Representative samples of the food source were periodically analyzed for pesticides, PCBs, and toxic metals by GeoLabs, Inc. and analyte levels were below LOC specified by ASTM (2005).	
Frequency of feeding:	2 times/day		
Details on feeding:	None.		

Table 3: Fish Selection and Pre-Exposure Performance.

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Age at test initiation:	*Ca.* 5 months	Ca. 20 weeks old	*EPA recommends reproductively mature (sexually dimorphic) fish, 4.5 - 6 months old.*

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Mean weight of males at test initiation (if determined):	3.2 g	Based on 20 males used to stock aquaria for pre-exposure phase.	*EPA recommends that a subsample of fish be weighed before the test to estimate the mean weight for each sex. It is recommended that the individual weight of each fish selected for the test be within ±20% of the estimated mean for each sex.*
Range of individual weights (males) at test initiation (if determined):	2.8 to 3.7	Individual weights within ±20% of the estimated mean.	
Mean weight of females at test initiation (if determined):	1.7 g	Based on 20 females used to stock aquaria for pre-exposure phase.	
Range of individual weights (females) at test initiation (if determined):	1.3 to 2.0	Individual weights within ±20% of the estimated mean.	
Mean length of males at test initiation (if determined):	Not reported		
Mean length of females at test initiation (if determined):	Not reported		
Duration of pre-exposure phase:	16 days		*EPA recommends a minimum of 14 days.*

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Were pre-exposure conditions identical to the definitive test?	Yes		*EPA recommends that pre-exposure conditions, including temperature, photoperiod, feeding, etc., be identical to definitive test conditions.*
Number of pre-exposure tanks:	36	Extra replicates were established to account for a potential lack of spawning in some chambers and/or mortality during this phase.	*EPA recommends that additional tanks set up at the beginning of pre-exposure will ensure that sufficient replicates with the correct sex ratio are available for the definitive test.*
Number of males per tank:	2		
Number of females per tank:	4		
Pre-exposure fecundity:	≥15 eggs/female/ reproductive day/ replicate		*EPA recommends that pre-exposure fecundity in each replicate (tank) selected for use in the definitive test be at least 15 eggs/female/reproductive day/replicate during the 7 days prior to the definitive test.*

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Number of spawns during pre-exposure:	≥two times in 7 days		*EPA recommends that spawning occur at least twice in the 7 days prior to the definitive test.*
Details on pre-exposure:		None.	

C. Exposure System

Table 4: Summary of Information on the Exposure System and Test Vessel Characteristics.

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Type of exposure:	Flow-through		*EPA recommends the use of a flow-through system. As noted in the Corrections and Clarifications document[1], the use of a static renewal system is not recommended for this assay.*
Type of flow-through dilution system:	Intermittent flow proportional diluters		*Intermittent flow proportional diluters or continuous flow serial diluters are recommended.[2]*

[1] U.S. Environmental Protection Agency (EPA). (2011). Corrections and Clarifications on Technical Aspects of the Test Guidelines for the Endocrine Disruptor Screening Program Tier 1 Assays (OCSPP Test Guideline Series 890). March 3, 2011. Office of Chemical Safety and Pollution Prevention (OCSPP), Washington, D.C. (http://www.epa.gov/endo/pubs/assayvalidation/clarificationdoc.pdf).

[2] Additional guidance for aquatic test design is located in OCSPP Guideline 850.1000, Special Considerations for Conducting Aquatic Laboratory Studies.

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Flow-through rate:	*Ca.* 45 mL/minute or 2.7 L/hr and 6.6 volume replacements/day.	Calculated by the reviewer based on 0.5 L/cycle, 131 cycles/24 hrs and 60 minutes/hr.	*Recommended flow-through rate is 45 mL/min (2.7 L/hr), or at least 6 total volume exchanges per day.*
Details on toxicant mixing for flow-through systems:		Flow-splitting chambers were used between diluter cells and the 4 replicate test vessels. Treatment recoveries were not measured prior to test initiation to indicate if diluter had reached equilibrium. Flow splitting accuracy not reported.	*Recommended toxicant mixing for flow-through systems: 1) Mixing chamber is recommended but not required; 2) Aeration is not recommended for mixing; 3) A demonstration that the test solution is completely mixed before introduced into the test system is recommended; 4) The recommended flow splitting accuracy is within 10%.*
Aeration?	None reported.		*EPA recommends aeration if dissolved oxygen reaches ≤4.9 mg/L (≤ 60% saturation).*

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Source of dilution water:	Well water		*EPA recommends natural or reconstituted water; it is recommended that natural water be sterilized with UV and tested for pesticides, heavy metals, and other possible contaminants. OECD accepts any water in which the test species show control survival at least as good as indicated in the test guideline.*
Was dilution water analyzed for pesticides, heavy metals, and other contaminants?	Yes		
Test vessel type/materials:	Glass aquaria with silicone adhesive.		*EPA and OECD recommend that water-contact portions of the system not compromise the study (e.g., all glass vessels or glass vessels with stainless steel frames are acceptable examples).*

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Test vessel size:	20 cm wide x 39 cm long x 25 cm high		*EPA recommends the use of 18 L test chambers (e.g., 40 x 20 x 20 cm).*
Fill volume:	10 L		*EPA recommends 10 L solution per tank.*
Spawning substrate material:	4 inch diameter aged PVC cut lengthwise in half (arches) and into 7 cm sections.		*EPA recommends that each tank contain three semi-circular spawning substrates, e.g., aged PVC pipe, 10 - 20 cm in length, split lengthwise.*
Spawning substrate size:	Aged PVC cut in half (arches) and into 7 cm sections.		
Additional details on exposure system:		None.	

Table 5: Summary of Water Quality Characteristics in the Test System.

Parameter	Minimum	Maximum	Mean	Measurement Interval	Guideline Recommendations
Temperature (°C)	25	26	25.5	Continuously	*EPA recommends temperature 25±1°C; inter-replicate and inter-treatment differentials should not exceed 1°C.*
pH	7.0	7.7	7.35	Weekly	*EPA recommends pH 6.5 to 9.0.*
Dissolved oxygen (mg/L)	4.9 (61%)	8.2 (98%)	6.6 (79.5%)	Weekly	*EPA recommends dissolved oxygen (DO) ≥4.9 mg/L (>60% air saturation)*
Total alkalinity (mg/L as $CaCO_3$)	20	26	23	Weekly	*EPA recommends total alkalinity >20 mg/L as $CaCO_3$.*
Hardness (mg/L as $CaCO_3$)	48	60	54	Weekly	
Total organic carbon (mg/L)	0.30	0.42	0.36	Twice	*EPA recommends that total organic carbon in dilution water be ≤2 mg/L.*
Unionized ammonia (µg/L)	Not reported	Not reported	Not reported	Not reported	*EPA recommends that unionized ammonia in the dilution water be ≤1 µg/L.*

Parameter	Minimum	Maximum	Mean	Measurement Interval	Guideline Recommendations
Residual chlorine (µg/L)	Not reported	Not reported	Not reported	Not reported	*EPA recommends that residual chlorine in dilution water be <10 µg/L.*
Conductivity (µmhos/cm)	260	340	300	Weekly	*General recommendations for frequency of measurements: EPA recommends that temperature, pH, and dissolved oxygen be measured in all test tanks at least weekly and that hardness and alkalinity be measured in controls and in one tank at the highest test concentration at least weekly. In addition, continuous temperature monitoring of at least one tank is encouraged.*

Abbreviations: [NA] Not applicable.

D. Study Design and Additional Experimental Conditions

Table 6: Range-Finding Study Conditions (if Applicable).

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Was a range-finder conducted?	Yes		*EPA recommends conducting a range-finder if 96-hour LC_{50} data for the fathead minnow are unavailable.*
If yes, what was the method for determining the highest test concentration in the range-finder?	14 day LC_{50} value range finding study.	Nominal test concentrations of 0 (negative and acetone controls), 0.00063, 0.0013, 0.0025, 0.0050 and 0.010 mg/L.	*EPA recommends that the highest test concentration be selected based on toxicity data for other fish studies or species, if available. Otherwise, either the solubility limit of the test compound or 100 mg/L (whichever is lower) is appropriate.*
Species:	*Pimephales promelas*		
Life stage:	Not reported		*EPA recommends that range-finding tests be performed with fish of similar age and size to those that would be utilized in the test.*

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Test duration:	14 day		*EPA recommends a 96-hour exposure.*
Additional details:	The test was conducted under flow-through conditions. On Days 12 and 13, two female fish at the highest treatment level were observed at solution's surface. At test termination, 40% combined male and female mortality was observed at the highest level. No other mortalities or sub-lethal effects were observed.		*EPA recommends conducting a range-finder with five test concentrations plus a control (six total treatment levels), with four females and two males per exposure tank (36 fish total). The number of mortalities that occur may be used to develop a concentration-response curve. Based upon the results, the highest concentration that does not result in increased mortality or signs of overt morbidity compared to controls, or 1/3 the derived 96-hr LC_{50} may be selected as the highest exposure concentration in the 21-day test.*

Table 7: Definitive Study Conditions.

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Test duration:	21 days		*EPA recommends that the duration of the definitive test be 21 days.*
Method for selecting the highest test concentration in the definitive test:	The toxicity 14 day LC_{50} value was estimated to be >0.010 mg/L.		*EPA recommends that the highest test concentration is either the solubility limit of the test compound, 100 mg/L, or demonstrates adequate evidence of toxicity (e.g., 1/3 the 96-hour LC_{50}), whichever concentration is lowest.*
Reference study citation (if applicable):	NA		
Separation of test concentrations:	0 (control), 0.00003, 0.0003, and 0.003 mg a.i./L	Step down factor of 10 from highest test concentration.	*EPA suggests that a concentration separation of between 0.33 (or three-fold) and 0.1 (or ten-fold) is scientifically acceptable*[1].

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Number of test concentrations:	4		*EPA recommends a minimum of 3 concentrations and a control, plus solvent control if appropriate.*
Are nominal concentrations adjusted for purity?	Yes		
Indicate the type of values presented for measured concentrations:	Mean measured		
Limit of quantification (LOQ):	<0.000001 mg a.i./L		*EPA recommends that for chemical test concentrations below the LOQ, analyses be conducted on the stock solutions.*
Level of detection (LOD):	Not reported		

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Frequency of measurement:	0, 4, 6, 11, and 19 days		*It is recommended that test item concentration be measured prior to the addition of fish in all tanks and at least weekly thereafter in two replicates per treatment level.*
Was the randomized complete block design used?	Yes		*EPA recommends that all fish be randomly assigned to tanks during pre-exposure. Tanks are then ranked according to pre-exposure fecundity, and the tanks with the highest fecundity are randomly assigned to a definitive test treatment and block first. Each block contains one replicate of each treatment, including controls.*
Number of replicates in control:	4		*EPA recommends 4 replicates.*
Number of replicates in solvent control (if applicable):	NA		*EPA recommends the use of a concurrent solvent control when a solubilizing agent is used. EPA recommends 4 replicates.*

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Number of replicates per test item treatment level:	4		*EPA recommends 4 replicates.*
Number of male fish per replicate at test initiation:	2		*EPA recommends 2 males per replicate.*
Number of female fish per replicate at test initiation:	4		*EPA recommends 4 females per replicate.*
Was a solvent used?	No		
Solvent type (if applicable):	NA		
Maximum solvent concentration (if applicable):	NA		*EPA recommends that the solvent not exceed 0.02 ml/L*[3]*. OECD recommends that solvent have no effect on survival nor produce any other adverse effects and that concentration not be greater than 0.1 ml/L*[4]*.*

[3] Hutchinson TH, Shillabeer N, Winter MJ, Pickford DB (2006). Acute and chronic effects of carrier solvents in aquatic organisms: A critical review. Review. Aquatic Toxicology, 76, pp.69–92.

[4] OECD (2000). Guidance Document on Aquatic Toxicity Testing of Difficult Substances and Mixtures. Environmental Health and Safety Publications. Series on

Parameter	Value(s)	Details or Remarks	Guideline Recommendations
Was a positive control used?	No		
Positive control (if applicable):	NA		
Positive control concentration(s) (if applicable):	NA		
Photoperiod:	16 hrs light: 8 hrs dark		*EPA recommends photoperiod 16:8 (light:dark).*
Light intensity at water's surface:	550-1000 lux	Fluorescent bulbs	*EPA recommends light intensity 540 – 1080 lux (at water's surface).*
Additional details:	None.		

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Table 8: Summary of Treatment Concentrations in the Fish Short-Term Reproduction Assay with Cypermethrin.

Treatment ID	Nominal Concentration (mg a.i./L)	Mean Measured Concentration[1] (mg a.i./L)	Mean CV (%)	Details or Remarks	Guideline Recommendations
Control (dilution water only)	<0.000001	<LOQ	NA		*EPA recommends that test item concentrations be maintained at a coefficient of variation (CV) ≤20%.*
Solvent control (if applicable)	NA	NA	NA		
Treatment 1	0.00003	0.000013	14.2	± 0.0.0016	
Treatment 2	0.0003	0.00012	45.6	± 0.0546	
Treatment 3	0.003	0.0014	51.2	± 0.7096	
Diluter Stock	NA	NA	NA		

Abbreviations: ^{CV} Coefficient of variation.

1 The mean measured values obtained by the study author are presented in the table and were calculated using the actual analytical results and not the rounded values presented. These values are different than the mean measured values obtained by the reviewer of 0.0000142, 0.00012 and 0.0014 mg a.i./L.

LOQ=0.000001 mg a.i./L.

E. Observations

Biological Endpoints: Survival, fecundity, fertilization success, and clinical signs were observed daily. At test termination (Day 21), secondary sex characterization (body color, pattern, body shape), body weight, length, tubercle score, gonadal staging and histopathology, and plasma vitellogenin were evaluated.

Were raw (individual) data provided? Yes

EPA recommends that observations of survival, fecundity, fertilization success, secondary sex characteristics, and other clinical signs occur at least daily. At test termination (Day 21), additional observations include body weight and length, nuptial tubercle score, gonadal staging and histopathology, plasma vitellogenin, and plasma sex steroids (testosterone and 17β-estradiol, if measured). Gonado-somatic index (GSI) is calculated using a ratio of gonad weight to body weight (gonad weight to the nearest 0.1 mg / body weight in mg x 100) at test termination.

Clinical signs of overt toxicity may include (but are not limited to) hemorrhage, cessation of feeding, and other abnormal behavior.

II. RESULTS AND DISCUSSION

A. Results

Mean male survival values were 100% at all test levels, and female survival values were 100, 94, 94, and 88% in the mean-measured 0 (control), 0.000013, 0.00012, and 0.0014 mg a.i./L treatment levels, respectively (Table 9). The mortality rate in the water control was 0%, therefore, the survival rate in the control group satisfied the minimum acceptable control value criteria of ≥90% according to the USEPA OCSPP 890.1350 guideline. During the in-life exposure, no notable observations occurred with regards to behavior, coloration/banding, changes in ovipositor appearance or size of dorsal nape pad.

Table 9: Adult Fish Survival in Fathead Minnow (*Pimephales promelas*).

Treatment (mg a.i./L) [mean-measured]	Males			Females		
	N[1]	# Surviving	% Survival	n	# Surviving	% Survival
Control (<LOQ)	9	9	100	15	15	100
0.000013	8	8	100	16	15	94
0.00012	8	8	100	16	15	94
0.0014	8	8	100	16	14	88

Abbreviations: [NA] Not applicable.

1 Total number of fish at test initiation.

LOQ=0.000001 mg a.i./L.

Mean male body weight values were 3.10, 3.57, 3.60, and 3.18 g and female body weight values were 1.45, 1.47, 1.26 and 1.28 g in the mean-measured 0 (control), 0.000013, 0.00012 and 0.0014 mg a.i./L treatment levels, respectively (Table 10). Mean male body length values were 62.2, 64.1, 64.3, and 62.7 mm and female body length values were 50.5, 49.5, 48.5 and 50.5 mm in the mean-measured 0 (control), 0.000013, 0.00012 and 0.0014 mg a.i./L treatment levels, respectively.

Table 10: Size at Test Termination in Fathead Minnow (*Pimephales promelas*).

Treatment (mg a.i./L) [mean-measured]	Body Weight						Length					
	Males			Females			Males			Females		
	n	Mean (g)	±SD	n	Mean (g)	±SD	n	Mean (mm)	±SD	n	Mean (mm)	±SD
Control (<LOQ)	4	3.10	0.39	4	1.45	0.50	4	62.2	2.44	4	50.5	2.99
0.000013	4	3.57	1.04	4	1.47	0.33	4	64.1	4.76	4	49.5	2.52
0.00012	4	3.60	1.24	4	1.26	0.24	4	64.3	5.89	4	48.5	2.98
0.0014	4	3.18	1.24	4	1.28	0.36	4	62.7	3.66	4	50.5	2.99

Abbreviations: [NA] Not applicable. [ND] Not determined. [SD] Standard deviation.

LOQ=0.000001 mg a.i./L.

Mean fecundity values were 14, 13, 8.3 and 2.2 eggs/female/day and fertilization success was 96, 97, 96 and 92% in the mean-measured 0 (control), 0.000013, 0.00012 and 0.0014 mg a.i./L treatment levels, respectively (Table 11). Replicate B of the control group inadvertently contained three male and three female fish due to a mis-sexed fish.

Table 11: Fecundity and Fertilization Success in Fathead Minnow (*Pimephales promelas*).

Treatment (mg a.i./L) [mean-measured]	Fecundity[1]	±SD	Fertilization Success (%)[2]	±SD
Control (<LOQ)	14	2.99	96	4.36
0.000013	13	3.00	97	0.5
0.00012	8.3	4.73	96	1.73
0.0014	2.2	0.88	92	6.65

Abbreviations: [NA] Not applicable. [ND] Not determined.

[1] Fecundity is calculated as the number of eggs per surviving female per reproductive day per replicate.

[2] Fertilization success (%) is calculated as the number of embryos divided by the number of eggs, multiplied by 100

LOQ=0.000001 mg a.i./L.

Median male tubercle scores were 32.5, 37, 33 and 25 in the mean-measured 0 (control), 0.000013, 0.00012 and 0.0014 mg a.i./L treatment levels, respectively (Table 12). None of the surviving females were found to have tubercles.

Table 12: Nuptial Tubercle Score in Fathead Minnow (*Pimephales promelas*).

Treatment (mg a.i./L) [mean-measured]	Males		Females	
	n	Median Tubercle Score	n	Median Tubercle Score
Control (<LOQ)	4	32.5	4	0
0.000013	4	37	4	0
0.00012	4	33	4	0
0.0014	4	25	4	0

Abbreviations: [NA] Not applicable. [ND] Not determined. [SD] Standard deviation.

LOQ=0.000001 mg a.i./L.

Median male GSI was 1.1, 1.2, 1.4 and 1.4% and mean female GSI was 12, 11, 11 and 14% in the mean-measured 0 (control), 0.000013, 0.00012 and 0.0014 mg a.i./L treatment levels, respectively (Table 13).

Table 13: Gonado-Somatic Index (GSI) in Fathead Minnow (*Pimephales promelas*).

Treatment (mg a.i./L) [mean-measured]	Males			Females		
	n	Median GSI[1] (%)	±SD	n	Mean GSI[1] (%)	±SD
Control (<LOQ)	4	1.1	0.25	4	12	4.3
0.000013	4	1.2	0.19	4	11	2.2
0.00012	4	1.4	0.07	4	11	2.4
0.0014	4	1.4	0.22	4	14	1.6

Abbreviations: [NA] Not applicable.

[1] Gonado-somatic index (%) is calculated as gonad weight (to the nearest 0.1 mg) / body weight (mg) x 100.

LQQ=0.000001 mg a.i./L.

Median male gonadal stage was 2, 2, 2 and 3 and median female gonadal stage was 3, 3, 3 and 3 in the mean-measured 0 (control), 0.000013, 0.00012 and 0.0014 mg a.i./L treatment levels, respectively (Table 14).

Table 14: Gonadal Staging in Fathead Minnow (*Pimephales promelas).*

Treatment (mg a.i./L) [mean-measured]	Males		Females	
	n	Median Stage[1]	n	Median Stage[2]
Control (<LOQ)	4	2	4	3
0.000013	4	2	4	3
0.00012	4	2	4	3
0.0014	4	3	4	3

Abbreviations: [J] Juvenile. [NA] Not applicable. [ND] Not determined. [UTS] Unable to stage.

[1] The guideline recommends the following gonadal staging scale for male fathead minnow: 0=undeveloped, 1=early spermatogenic, 2=mid-spermatogenic, 3=late spermatogenic, 4=spent.

[2] The guideline recommends the following gonadal staging scale for female fathead minnow: 0=undeveloped, 1=early development, 2=mid-development, 3=late development, 4=late development/hydrated, 5=post-ovulatory.

LOQ=0.000001 mg a.i./L.

In male fish, there were only background or sporadic findings noted and were not related to treatment (Tables 15-16). In female fish, there was an increased incidence of mature oocyte atresia at the 0.0014 mg a.i./L treatment, which was associated with granulomatous inflammation and the presence of microsporidia in half the observed cases across all treatment levels (Tables 17-18).

Table 15: Gonadal Histopathology in Male Fathead Minnow (*Pimephales promelas*).

Treatment (mg a.i./L) [mean-measured]	Severity	Diagnostic Observations[1]									
		Increased Proportion of Spermatogonia		Presence of Testis-Ova		Increased Testicular Degeneration		Duct Mineralization		Interstitial cell (Leydig) hypertrophy/hyperplasia	
		n	Incidence	n	Incidence	n	Incidence	n	Incidence	n	Incidence
Control (<LOQ)	0	9	7	9	8	9	8	NA	NA	NA	NA
	1	9	1	9	1	9	1	NA	NA	NA	NA
	2	9	1	9	0	9	0	NA	NA	NA	NA
	3	9	0	9	0	9	0	NA	NA	NA	NA
	4	9	0	9	0	9	0	NA	NA	NA	NA
0.000013	0	8	8	8	8	8	7	NA	NA	NA	NA
	1	8	0	8	0	8	0	NA	NA	NA	NA
	2	8	0	8	0	8	1	NA	NA	NA	NA
	3	8	0	8	0	8	0	NA	NA	NA	NA
	4	8	0	8	0	8	0	NA	NA	NA	NA
0.00012	0	8	6	8	8	8	8	NA	NA	NA	NA
	1	8	1	8	0	8	0	NA	NA	NA	NA
	2	8	1	8	0	8	0	NA	NA	NA	NA
	3	8	0	8	0	8	0	NA	NA	NA	NA
	4	8	0	8	0	8	0	NA	NA	NA	NA
0.0014	0	8	8	8	8	8	8	NA	NA	NA	NA
	1	8	0	8	0	8	0	NA	NA	NA	NA
	2	8	0	8	0	8	0	NA	NA	NA	NA
	3	8	0	8	0	8	0	NA	NA	NA	NA
	4	8	0	8	0	8	0	NA	NA	NA	NA

[1] Gonadal histopathology diagnostic observations are graded 0 – 4 based on severity: 0=Not remarkable, 1=Minimal, 2=Mild, 3=Moderate, 4=Severe. See Appendix E of the test guideline for reference.

LOQ=0.000001 mg a.i./L.

Table 16: Additional Gonadal Histopathology Observations in Male Fathead Minnow (*Pimephales promelas*).

Treatment (mg a.i./L) [mean-measured]	Severity	Additional Diagnostic Observations[1]									
		Decreased Proportion of Spermatogonia		Increased Vascular or Interstitial Proteinaceous Fluid		Asynchronous Gonad Development		Altered Proportions of Spermatocytes or Spermatids		Granulomatous Inflammation	
		n	Incidence	n	Incidence	n	Incidence	n	Incidence	n	Incidence
Control (<LOQ)	0	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	1	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	2	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	3	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	4	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
0.000013	0	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	1	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	2	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	3	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	4	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA

Treatment (mg a.i./L) [mean-measured]	Severity	Additional Diagnostic Observations[1]									
		Decreased Proportion of Spermatogonia		Increased Vascular or Interstitial Proteinaceous Fluid		Asynchronous Gonad Development		Altered Proportions of Spermatocytes or Spermatids		Granulomatous Inflammation	
		n	Incidence	n	Incidence	n	Incidence	n	Incidence	n	Incidence
0.00012	0	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	1	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	2	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	3	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	4	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
0.0014	0	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	1	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	2	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	3	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
	4	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA

[1] Gonadal histopathology diagnostic observations are graded 0 – 4 based on severity: 0=Not remarkable, 1=Minimal, 2=Mild, 3=Moderate, 4=Severe. See Appendix E of the test guideline for reference.

LOQ=0.000001 mg a.i./L.

Table 17: Gonadal Histopathology in Female Fathead Minnow (*Pimephales promelas*).

Treatment (mg a.i./L) [mean-measured]	Additional Diagnostic Observations[1]								
	Severity	Increased Oocyte Atresia		Perifollicular Cell Hyperplasia/ Hypertrophy		Decreased Yolk Formation		Aggregates of macrophages, multifocal	
		n	Incidence	n	Incidence	n	Incidence	n	Incidence
Control (<LOQ)	0	15	15	NA	NA	NA	NA	NA	NA
	1	15	0	NA	NA	NA	NA	NA	NA
	2	15	0	NA	NA	NA	NA	NA	NA
	3	15	0	NA	NA	NA	NA	NA	NA
	4	15	0	NA	NA	NA	NA	NA	NA
0.000013	0	15	13	NA	NA	NA	NA	NA	NA
	1	15	2	NA	NA	NA	NA	NA	NA
	2	15	0	NA	NA	NA	NA	NA	NA
	3	15	0	NA	NA	NA	NA	NA	NA
	4	15	0	NA	NA	NA	NA	NA	NA
0.00012	0	15	14	NA	NA	NA	NA	NA	NA
	1	15	1	NA	NA	NA	NA	NA	NA
	2	15	0	NA	NA	NA	NA	NA	NA
	3	15	0	NA	NA	NA	NA	NA	NA
	4	15	0	NA	NA	NA	NA	NA	NA

Treatment (mg a.i./L) [mean-measured]	Additional Diagnostic Observations[1]								
	Severity	Increased Oocyte Atresia		Perifollicular Cell Hyperplasia/ Hypertrophy		Decreased Yolk Formation		Aggregates of macrophages, multifocal	
		n	Incidence	n	Incidence	n	Incidence	n	Incidence
0.0014	0	14	7	NA	NA	NA	NA	NA	NA
	1	14	1	NA	NA	NA	NA	NA	NA
	2	14	3	NA	NA	NA	NA	NA	NA
	3	14	3	NA	NA	NA	NA	NA	NA
	4	14	0	NA	NA	NA	NA	NA	NA

[1] Gonadal histopathology diagnostic observations are graded 0 – 4 based on severity: 0=Not remarkable, 1=Minimal, 2=Mild, 3=Moderate, 4=Severe. See Appendix E of the test guideline for reference.

LOQ=0.000001 mg a.i./L.

Table 18: Additional Gonadal Histopathology Observations in Female Fathead Minnow (*Pimephales promelas*).

Treatment (mg a.i./L) [mean-measured]	Additional Diagnostic Observations[1]								
	Severity	Interstitial fibrosis		Egg Debris in Oviduct		Granulomatous Inflammation		Decreased Post-Ovulatory Follicles	
		n	Incidence	n	Incidence	n	Incidence	n	Incidence
Control (<LOQ)	0	NA	NA	15	13	15	14	NA	NA
	1	NA	NA	15	1	15	1	NA	NA
	2	NA	NA	15	1	15	0	NA	NA
	3	NA	NA	15	0	15	0	NA	NA
	4	NA	NA	15	0	15	0	NA	NA

Treatment (mg a.i./L) [mean-measured]	Additional Diagnostic Observations[1]								
	Severity	Interstitial fibrosis		Egg Debris in Oviduct		Granulomatous Inflammation		Decreased Post-Ovulatory Follicles	
		n	Incidence	n	Incidence	n	Incidence	n	Incidence
0.000013	0	NA	NA	15	15	15	13	NA	NA
	1	NA	NA	15	0	15	2	NA	NA
	2	NA	NA	15	0	15	0	NA	NA
	3	NA	NA	15	0	15	0	NA	NA
	4	NA	NA	15	0	15	0	NA	NA
0.00012	0	NA	NA	15	15	15	13	NA	NA
	1	NA	NA	15	0	15	2	NA	NA
	2	NA	NA	15	0	15	0	NA	NA
	3	NA	NA	15	0	15	0	NA	NA
	4	NA	NA	15	0	15	0	NA	NA
0.0014	0	NA	NA	14	13	14	11	NA	NA
	1	NA	NA	14	0	14	0	NA	NA
	2	NA	NA	14	1	14	3	NA	NA
	3	NA	NA	14	0	14	0	NA	NA
	4	NA	NA	14	0	14	0	NA	NA

[1] Gonadal histopathology diagnostic observations are graded 0 – 4 based on severity: 0=Not remarkable, 1=Minimal, 2=Mild, 3=Moderate, 4=Severe. See Appendix E of the test guideline for reference.

LOQ=0.000001 mg a.i./L.

Mean male VTG was 41, 53, 39 and 35 and female VTG was $1.2x10^6$, $8.4x10^5$, $1.1x10^6$ and $1.1x10^6$ ng/mL in the mean-measured 0 (control), 0.000013, 0.00012 and 0.0014 mg a.i./L treatment levels, respectively (Table 19).

Table 19: Plasma Vitellogenin in Fathead Minnow (*Pimephales promelas*).

Treatment (mg a.i./L) [mean-measured]	Plasma Vitellogenin (VTG)					
	Males			Females		
	n	Mean (ng/mL plasma)	±SD	n	Mean (ng/mL plasma)	±SD
Control (<LOQ)	4	41	34	4	1.2×10^6	2.553×10^5
0.000013	4	53	72	4	8.4×10^5	2.34×10^5
0.00012	4	39	25	4	1.1×10^6	3.64×10^5
0.0014	4	35	29	4	1.1×10^6	3.01×10^5

Abbreviations: NA Not applicable. ND Not determined. SD Standard deviation.

LOQ=0.000001 mg a.i./L.

Mean plasma testosterone and plasma 17β-estradiol in male and females was not measured (Table 20).

Table 20: Plasma Sex Steroids in Fathead Minnow (*Pimephales promelas*). Not measured.

Treatment (mg a.i./L) [mean-measured]	Plasma Testosterone (T)						Plasma 17β-estradiol (E2)					
	Males			Females			Males			Females		
	n	Mean (ng/mL plasma)	±SD	n	Mean (ng/mL plasma)	±SD	n	Mean (ng/mL plasma)	±SD	n	Mean (ng/mL plasma)	±SD
Control (<LOQ)	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
0.000013	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
0.00012	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
0.0014	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA

Abbreviations: [NA] Not applicable. [ND] Not determined. [SD] Standard deviation.

LOQ=0.000001 mg a.i./L.

There were no notable observations with regards to behavior or changes in appearance (coloration/banding, presence of ovipositor and dorsal nape pad), including secondary sex characteristics in the control or treated groups (Table 21). Replicate B in the control group contained three males and three females due to mis-sexed fish.

Table 21: Secondary Sex Characteristics and Clinical Signs in Fathead Minnow (*Pimephales promelas*).

Treatment (mg a.i./L) [mean-measured]	Secondary Sex Characteristics and Clinical Signs					
	Males			Females		
	Type	n	Incidence	Type	n	Incidence
Control (<LOQ)	None	9	0	None	15	0
0.000013	None	8	0	None	15	0
0.00012	None	8	0	None	15	0
0.0014	None	8	0	None	14	0

LOQ=0.000001 mg a.i./L.

B. Study Author's Analysis and Conclusions

The study author analyzed tubercle score, GSI, fertility, fecundity, and VTG. Data were gender specific and analyzed in comparison to the controls.

Descriptive statistics (mean, standard deviation, etc.) were determined for each endpoint. Significant effects were detected for $p<0.05$ with the exception of Shapiro-Wilk's and Bartlett's Tests, which were based on $p<0.01$ (CETIS, version 1.8.4, 2011). Survival, body weight and body length data were recoded but not statistically analyzed. Fecundity, fertility, male and female VTG, and female GSI were analyzed using ANOVA and a one-tailed Dunnett's Multiple Comparison test. Median male tubercle scores and median male GSI were analyzed by ANOVA and Jonckheere-Terpstra Step-Down test. Male and female length was recorded but statistical analysis was not reported. Prior to Dunnett's, data were analyzed by Shapiro-Wilk's test and Bartlett's to test for normality and homogeneity of variances, respectively. If normality and homogeneity tests passed ($p>0.01$), a parametric analysis was performed using the transformed data. If non-normality or unequal variance were indicated ($p<0.01$), a non-parametric analysis was performed on the ranks of the data. These methods appear to be consistent with the methods recommended in the guideline.

There were no differences in male or female survival at any treatment level based on visual inspection of the data (Table 9). There was no statistically significant differenca in fertilization success at any treatment level compared to the control ($p>0.05$; Dunnett's). There was a statistically significant reduction in fecundity at the 0.00012 and 0.0014 mg a.i./L treatment levels compared to the control ($p<0.05$; Dunnett's). However, the mean fecundity in the control was 14 eggs/female/day; OCSPP guidance requires fecundity in the controls of at least 15 eggs/female/day. There was a significant decrease in median male GSI at the 0.00012 and 0.0014 mg a.i./L treatment levels compared to the control ($p<0.05$; Jonckheere-Terpstra Step-Down test; Table 13). There were no significent differences between treated groups and control for any other endpoint.

In male fish, there were only background or sporadic gonadal histopathology findings noted which were not related to treatment (Tables 15-16). In female fish, there was an increased incidence of

mature oocyte atresia at the 0.0014 mg a.i./L treatment, which was associated with granulomatous inflammation and the presence of microsporidia in half the observed cases across all treatment levels (Tables 17-18). There were no significant findings observed in the gonadal stage for either males or females.

C. Reviewer's Analysis and Conclusions

Statistical Methods: The reviewer analyzed survival (mortality) data based on visual observation. Male GSI and fecundity were consistent with a monotonic concentration-response; no other endpoints were associated with a monotonic response. All data ware tested for normality using Shapiro-Wilks test and for homogeneity of variance using Levene's test. Data which met the assumptions of normality and homogeneity of variance were then analyzed using the parametric Dunnett's test. Data which did not meet the parametric assumptions were analyzed using the non-parametric Mann-Whitney U test. Male GSI and fecundity exhibited monotonic trends and was analyzed using the non-parametric Jonckheere-Terpstra test, as recommended by the OCSPP 890.1350 guideline.

None of the surviving females were found to have tubercles. Unless otherwise indiceted, effects were considered statistically significant at $p<0.05$. These analyses were conducted using SAS® (SAS Institute, Cary, NC; version 8.1). The data was subsequently re-run for the gender-specific endpoints (male GSI, male VTG, male length, male weight) when excluding the mis-sexed male fish in the control group with CETIS (Version 1.8.7.12). Both the study author and reviewer relied on the mean-measured concentrations to discuss effects in this study.

Conclusions:

There were no significant differences in either male or female survival at any treatment levels compared to the negative control based on visual observation (Table 9). Male GSI exhibited a significant ($p<0.05$) increase of 22 to 27% at the 0.00012 and 0.0014 mg a.i./L treatment levels compared to the negative control. Fecundity was significantly ($p<0.05$) reduced 42% and 84% of control at the 0.00012 and 0.0014 mg a.i./L treatment level. There were no other significant

effects on all other endpoints (female GSI, male and female plasma VTG, male and female weight and length and male and female nuptial tubercle score).

Although not analyzed statistically, in male fish, there were only background or sporadic findings noted and were not related to treatment. In female fish, there was an increased incidence of mature oocyte atresia at the 0.0014 mg a.i./L treatment, which was associated with granulomatous inflammation and the presence of microsporidia in half the observed cases across all treatment levels.

There were no significant findings observed in the gonadal stage for either males or females, and no notable observations in secondary sex characteristics or clinical signs for any treatment group compared to the controls. Sex steroids were not measured in this study.

Table 22: Reproductive and HPG Endpoints[1,2] for Male Fathead Minnow (*Pimephales promelas)* in the FSTRA with Cypermethrin.

Treatment (mg a.i./L) [mean-measured]	Tubercle Score		GSI		Gonadal Staging and Histo.	Plasma VTG		Plasma T		Plasma E2	
	Median	p	% Diff.	p	Effect? (Yes/No)	% Diff.	P	% Diff.	p	% Diff.	p
Control (<LOQ)	32.5	NA	0	NA	No	0	NA	NA	NA	NA	NA
0.000013	37	0.914	10.41	0.231	No	59.4	0.859	NA	NA	NA	NA
0.00012	33	0.600	22.17	0.041	No	18.05	0.994	NA	NA	NA	NA
0.0014	25	0.071	26.70	0.018	No	5.26	1.00	NA	NA	NA	NA
Statistical Test	Mann-Whitney		Jonckheere's		NA	Dunnett's		NA		NA	

Abbreviations: Conc. Concentration. Diff. Difference. E2 17β-estradiol. GSI Gonado-Somatic Index. Histo. Histopathology. NA Not applicable. T Testosterone. VTG Vitellogenin. NA Not applicable.

1 Unless otherwise indicated, effects and percent (%) differences are reported based on comparison to the negative (clean water) control. Conclusions regarding histopathology may be heavily weighted by the expert opinion of a board-certified pathologist.

2 Unless otherwise specified, effects are considered statistically significant at $p<0.05$.

LOQ=0.000001 mg a.i./L.

Table 23: Reproductive and HPG Endpoints[1,2] for Female Fathead Minnow (*Pimephales promelas)* in the FSTRA with Cypermethrin.

Treatment (mg a.i./L) [mean-measured]	Fecundity		Fert. Success		Tubercle Score		GSI		Gonadal Staging and Histo.	Plasma VTG		Plasma T		Plasma E2	
	% Diff.	p	% Diff.	P	Median	p	% Diff.	p	Effect? (Yes/No)	% Diff.	p	% Diff.	p	% Diff.	p
Control (<LOQ)	0	NA	0	NA	0	NA	0	NA	No	0	NA	NA	NA	NA	NA
0.000013	-5.26	0.328	1.30	0.9140	0	NA	-5.05	0.979	No	-28.30	0.302	NA	NA	NA	NA
0.00012	-41.93	0.032	0.00	>0.05	0	NA	-5.68	0.971	No	-5.32	0.981	NA	NA	NA	NA
0.0014	-84.39	<0.001	-4.40	0.659	0	NA	20.00	0.492	No	-10.64	0.879	NA	NA	NA	NA
Statistical Test	Jonckheere's		Dunnett's T3		NA		Dunnett's		NA	Dunnett's		NA		NA	

Abbreviations: Conc. Concentration. Diff. Difference. E2 17β-estrediol. Fert. Fertilization. GSI Gonado-Somatic Index. Histo. Histopathology.

NA Not applicable. T Testosterone. VTG Vitellogenin.

[1] Unless otherwise indicated, effects and percent (%) differences are reported based on comparison to the negative (clean water) control. Conclusions regarding histopathology may be heavily weighted by the expert opinion of a board-certified pathologist.

[2] Unless otherwise specified, effects are considered statistically significant at $p<0.05$.

LOQ=0.000001 mg a.i./L.

Table 24: Growth Endpoints[1,2] in the Fish Short-Term Reproduction Assay (FSTRA) with Cypermethrin.

Treatment (mg a.i./L) [mean-measured]	Body Weight				Length			
	Males		Females		Males		Females	
	% Diff.	p	% Diff.	p	% Diff.	p	% Diff.	p
Control (<LOQ)	0	NA	0	NA	0	NA	0	NA
0.000013	15.22	0.497	5.83	0.808	2.97	0.651	-2.37	0.502
0.00012	16.16	0.435	-9.03	0.552	3.32	0.577	-4.43	0.102
0.0014	2.64	0.993	-9.29	0.531	0.75	0.989	-1.55	0.768
Statistical Test	Dunnett's		Dunnett's		Dunnett's		Dunnett's	

Abbreviations: Diff. Difference. NA Not applicable.

[1] Unless otherwise indicated, percent (%) differences are reported based on comparison to the negative (clean water) control.

[2] Unless otherwise specified, effects are considered statistically significant at $p<0.05$.

LOQ=0.000001 mg a.i./L.

E. Study Deficiencies

There were deviations in the validity and performance criteria and a couple of minor deviations described in Section I. Materials and Methods of this DER regarding failure to report certain water characteristics and meet environmental criteria:

1. There were one replicate in the control group (Replicate C) where there was a period of 4 days in which no spawning occurred, and fecundity was 13 eggs/female/day, respectively. Therefore, this replicate did not meet the criteria of either at least 15 eggs/female/day or spawning at least every 4 days. However, it is noted that the overall control mean fecundity value (14) was just slightly below 15 eggs/female/day, and the

other two replicates did achieve 15 or greater eggs/female/day and the remaining replicate spawned at least every 4 days (11 eggs/female/day).

2. Analytical verification of the test material from Days 0, 6, 11, 19 and 21 yielded recoveries of 44% to 67% of nominal concentrations. Additionally, the %CV values were 14.2, 45.6 and 51.2%, respectively, and did not meet the validation criteria of <20%. Results of these analyses indicate measured concentrations were generally lower than nominal, but according to the study author were consistent with expectations for the physicochemical properties and previous experience with the test item. Therefore, although there was no pattern of decline during the study period and no un-dissolved test substance was observed in the dilution system, the recoveries indicate the test material was generally poorly recovered in solution under the test conditions.

3. The test material was detected at Days 11 (one replicate) and 19 (two replicates) in the control samples. The study author concluded that the contamination was not consistent among replicates and not consistent throughout exposure. The study author concluded the contamination was likely a result of processing samples after being removed from the exposure system, indicating the control fish were not exposed to cypermethrin.

4. The unionized ammonia and residual chlorine in the test water were not reported. The OCSPP 890.1350 performance criteria establish maximum levels for these values and it is unclear if the maximum recommendations were exceeded.

5. Incorrect sexing of individuals in one replicate from the control group resulted in there being three males and three females in this replicate instead of the two males and four females specified in the OCSPP 890.1350 guideline.

These deviations did not sufficiently adversely impact interpretation of the assay results. The remaining validity and performance criteria for OCSPP 890.1350 were met.

F. Reviewer's Comments

The reviewer's and the study author's results were in agreement. Both the study author's and reviewer's analysis detected a statistically significant reduction ($p<0.05$) in fecundity and a statistically significant ($p<0.05$) increase in male GSI score at the 0.00012 and 0.0014 mg a.i./L treatment levels compared to the negative control. The reviewer's conclusions based on the OCSPP 890.1350 flowchart are presented in the Executive Summary and Conclusions sections of this DER.

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APPENDIX I. OUTPUT OF REVIEWER'S STATISTICAL VERIFICATION:

test for fish screen study - Cypermethrin
ANALYSIS RESULTS FOR VARIABLE VAR01 (F body weight (g))

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS
Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01
Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05
Use parametric analyses if neither test rejected, otherwise non-parametric analyses.

Shapiro-Wilks Test Stat	Shapiro-Wilks P-value	Levenes Test Stat	Levenes P-value	Conclusion
0.913	0.132	1.115	0.381	USE PARAMETRIC TESTS

****

BASIC SUMMARY STATISTICS

Level	N	Mean	StdDev	StdErr	Coef of Var	95% Conf.	Interval
Ctrl	4	1.39	0.20	0.10	14.28	1.08,	1.71
Dose1	4	1.48	0.13	0.07	8.84	1.27,	1.68
Dose2	4	1.27	0.11	0.05	8.29	1.10,	1.44
Dose3	4	1.26	0.18	0.09	13.92	0.98,	1.54

Level	Median	Min	Max	%of Control(means)	%Reduction(means)
Ctrl	1.32	1.25	1.69	.	.
Dose1	1.47	1.34	1.63	105.83	-5.83
Dose2	1.24	1.18	1.41	90.97	9.03
Dose3	1.27	1.09	1.44	90.71	9.29

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	1.71	0.218

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing negative trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	1.39	.	1.43	.	.	.	.	.	.
Dose1	1.48	0.808	1.43	0.727	.	.	.	.	.
Dose2	1.27	0.552	1.27	0.177	0.292	.	.	.	.
Dose3	1.26	0.531	1.26	0.175	0.279	1.000	.	.	.

****

NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	3.86	0.277

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing negative trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	1.32	.	.
Dose1	1.47	0.494	0.807
Dose2	1.24	0.346	0.153
Dose3	1.27	0.678	0.081

DECREASING TREND TEST SUMMARY	LOWEST CONCENTRATION SIGNIF. LESS THAN CONTROL
Williams	>highest dose (no sign. differences)
Jonckheere	>highest dose (no sign. differences)

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	1.71	0.218

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing INCREASING trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	-1.39	.	-1.35	.	.	.	.	.	.
Dose1	-1.48	0.808	-1.35	0.737	.	.	.	.	.
Dose2	-1.27	0.552	-1.35	0.770	0.292	.	.	.	.
Dose3	-1.26	0.531	-1.35	0.788	0.279	1.000	.	.	.

****

NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	3.86	0.277

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing INCREASING trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	-1.32	.	.
Dose1	-1.47	0.494	0.193
Dose2	-1.24	0.346	0.847
Dose3	-1.27	0.678	0.919

INCREASING TREND TEST SUMMARY	LOWEST CONCENTRATION SIGNIF. GREATER THAN CONTROL
Williams	>highest dose (no sign. differences)
Jonckheere	>highest dose (no sign. differences)

test for fish screen study - Cypermethrin
ANALYSIS RESULTS FOR VARIABLE VAR02 (M body weight (g))

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS
Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01
Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05
Use parametric analyses if neither test rejected, otherwise non-parametric analyses.

Shapiro-Wilks Test Stat	Shapiro-Wilks P-value	Levenes Test Stat	Levenes P-value	Conclusion
0.936	0.305	3.162	0.064	USE PARAMETRIC TESTS

****

BASIC SUMMARY STATISTICS

Level	N	Mean	StdDev	StdErr	Coef of Var	95% Conf.Interval	
Ctrl	4	3.02	0.37	0.19	12.34	2.43,	3.61
Dose1	4	3.57	0.78	0.39	21.91	2.33,	4.81
Dose2	4	3.60	0.62	0.31	17.10	2.62,	4.58
Dose3	4	3.18	0.17	0.09	5.41	2.91,	3.45

Level	Median	Min	Max	%of Control(means)	%Reduction(means)
Ctrl	3.00	2.64	3.44	.	.
Dose1	3.84	2.42	4.17	118.17	-18.17
Dose2	3.63	2.93	4.20	119.13	-19.13
Dose3	3.19	2.97	3.38	105.26	-5.26

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	1.14	0.373

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing negative trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	3.02	.	3.40	.	.	.	.	.	.
Dose1	3.57	0.374	3.40	0.895	.	.	.	.	.
Dose2	3.60	0.337	3.40	0.917	1.000	.	.	.	.
Dose3	3.18	0.953	3.18	0.796	0.739	0.696	.	.	.

****

NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	3.04	0.385

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing negative trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	3.00	.	.
Dose1	3.84	0.346	0.876
Dose2	3.63	0.235	0.928

```
  Dose3      3.19                    0.678                    0.680

 DECREASING TREND TEST SUMMARY      LOWEST CONCENTRATION SIGNIF. LESS THAN CONTROL
   Williams                                >highest dose (no sign. differences)
   Jonckheere                              >highest dose (no sign. differences)

*****************************************************************************
PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
    Analysis of Variance (ANOVA) - overall F-test
     Numerator df    Denominator df    F-stat       P-value
           3              12              1.14        0.373

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing INCREASING trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level     Mean    Dunnett  Isotonic  Williams                Tukey p-values
                  p-value    mean     p-value    Dose1   Dose2   Dose3   Dose4   Dose5

 Ctrl     -3.02     .        -3.02       .          .       .       .       .       .
 Dose1    -3.57   0.374      -3.45     0.168        .       .       .       .       .
 Dose2    -3.60   0.337      -3.45     0.179      1.000     .       .       .       .
 Dose3    -3.18   0.953      -3.45     0.186      0.739   0.696     .       .       .

*****************************************************************************
NON-PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
    Kruskal-Wallis test - equality among treatment groups
     Degrees of Freedom   TestStat    P-value
           3                3.04       0.385

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing INCREASING trend

 Level     Median          MannWhit p-value          Jonckheere p-value
  Ctrl      -3.00                    .                        .
  Dose1     -3.84                  0.346                    0.124
  Dose2     -3.63                  0.235                    0.072
  Dose3     -3.19                  0.678                    0.320

 INCREASING TREND TEST SUMMARY      LOWEST CONCENTRATION SIGNIF. GREATER THAN CONTROL
   Williams                                >highest dose (no sign. differences)
   Jonckheere                              >highest dose (no sign. differences)

test for fish screen study - Cypermethrin
ANALYSIS RESULTS FOR VARIABLE VAR03  ( F body length (mm) )

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS
Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01
Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05
Use parametric analyses if neither test rejected, otherwise non-parametric analyses.
  Shapiro-Wilks  Shapiro-Wilks    Levenes    Levenes   Conclusion
    Test Stat      P-value       Test Stat   P-value
      0.912         0.126          1.347      0.306    USE PARAMETRIC TESTS
```

****

BASIC SUMMARY STATISTICS

Level	N	Mean	StdDev	StdErr	Coef of Var	95% Conf.	Interval
Ctrl	4	50.81	0.98	0.49	1.93	49.25,	52.37
Dose1	4	49.61	1.15	0.57	2.32	47.78,	51.44
Dose2	4	48.56	0.91	0.46	1.88	47.11,	50.01
Dose3	4	50.02	2.18	1.09	4.36	46.56,	53.49

Level	Median	Min	Max	%of Control(means)	%Reduction(means)
Ctrl	50.72	49.72	52.10	.	.
Dose1	49.58	48.45	50.83	97.63	2.37
Dose2	48.17	47.99	49.92	95.57	4.43
Dose3	50.83	46.89	51.54	98.45	1.55

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	1.79	0.202

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing negative trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	50.81	.	50.81	.	.	.	.	.	.
Dose1	49.61	0.502	49.61	0.148	.	.	.	.	.
Dose2	48.56	0.102	49.29	0.094	0.719	.	.	.	.
Dose3	50.02	0.768	49.29	0.097	0.975	0.480	.	.	.

****

NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	5.05	0.168

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing negative trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	50.72	.	.
Dose1	49.58	0.346	0.124
Dose2	48.17	0.103	0.006
Dose3	50.83	0.889	0.131

DECREASING TREND TEST SUMMARY — LOWEST CONCENTRATION SIGNIF. LESS THAN CONTROL

Williams >highest dose (no sign. differences)

Jonckheere >highest dose (no sign. differences)

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	1.79	0.202

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing INCREASING trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	-50.81	.	-49.66	.	.	.	.	.	.
Dose1	-49.61	0.502	-49.66	0.923	.	.	.	.	.
Dose2	-48.56	0.102	-49.66	0.941	0.719	.	.	.	.
Dose3	-50.02	0.768	-50.02	0.895	0.975	0.480	.	.	.

****
NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	5.05	0.168

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing INCREASING trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	-50.72	.	.
Dose1	-49.58	0.346	0.876
Dose2	-48.17	0.103	0.994
Dose3	-50.83	0.889	0.869

INCREASING TREND TEST SUMMARY	LOWEST CONCENTRATION SIGNIF. GREATER THAN CONTROL
Williams	>highest dose (no sign. differences)
Jonckheere	>highest dose (no sign. differences)

test for fish screen study - Cypermethrin
ANALYSIS RESULTS FOR VARIABLE VAR04 (M body length (mm))

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS
Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01
Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05
Use parametric analyses if neither test rejected, otherwise non-parametric analyses.

Shapiro-Wilks Test Stat	Shapiro-Wilks P-value	Levenes Test Stat	Levenes P-value	Conclusion
0.910	0.118	0.652	0.597	USE PARAMETRIC TESTS

****
BASIC SUMMARY STATISTICS

Level	N	Mean	StdDev	StdErr	Coef of Var	95% Conf.Interval
Ctrl	4	61.64	2.27	1.13	3.68	58.03, 65.25
Dose1	4	64.07	3.62	1.81	5.65	58.31, 69.82
Dose2	4	64.28	2.64	1.32	4.10	60.08, 68.48

Dose3	4	62.68	1.69	0.84	2.69	60.00, 65.37

Level	Median	Min	Max	%of Control(means)	%Reduction(means)
Ctrl	61.78	58.82	64.19	.	.
Dose1	65.35	58.93	66.63	103.93	-3.93
Dose2	64.89	60.91	66.43	104.28	-4.28
Dose3	62.24	61.31	64.94	101.69	-1.69

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	0.88	0.481

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing negative trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	61.64	.	63.33	.	.	.	.	.	.
Dose1	64.07	0.455	63.33	0.878	.	.	.	.	.
Dose2	64.28	0.392	63.33	0.902	0.999	.	.	.	.
Dose3	62.68	0.900	62.68	0.838	0.880	0.828	.	.	.

****

NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	2.60	0.457

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing negative trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	61.78	.	.
Dose1	65.35	0.346	0.876
Dose2	64.89	0.346	0.810
Dose3	62.24	0.494	0.610

DECREASING TREND TEST SUMMARY — LOWEST CONCENTRATION SIGNIF. LESS THAN CONTROL

Williams >highest dose (no sign. differences)
Jonckheere >highest dose (no sign. differences)

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	0.88	0.481

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing INCREASING trend

Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	-61.64	.	-61.64	.	.	.	.	.	.
Dose1	-64.07	0.455	-63.68	0.178	.	.	.	.	.
Dose2	-64.28	0.392	-63.68	0.190	0.999	.	.	.	.
Dose3	-62.68	0.900	-63.68	0.197	0.880	0.828	.	.	.

****

NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	2.60	0.457

MannWhit - testing each trt median signif. different from control

Jonckheere - test assumes dose-response relationship, testing INCREASING trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	-61.78	.	.
Dose1	-65.35	0.346	0.124
Dose2	-64.89	0.346	0.190
Dose3	-62.24	0.494	0.390

INCREASING TREND TEST SUMMARY	LOWEST CONCENTRATION SIGNIF. GREATER THAN CONTROL
Williams	>highest dose (no sign. differences)
Jonckheere	>highest dose (no sign. differences)

test for fish screen study - Cypermethrin

ANALYSIS RESULTS FOR VARIABLE VAR05 (F vitellogenin (ng/mL))

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS

Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01

Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05

Use parametric analyses if neither test rejected, otherwise non-parametric analyses.

Shapiro-Wilks Test Stat	Shapiro-Wilks P-value	Levenes Test Stat	Levenes P-value	Conclusion
0.941	0.361	0.460	0.715	USE PARAMETRIC TESTS

****

BASIC SUMMARY STATISTICS

Level	N	Mean	StdDev	StdErr	Coef of Var	95% Conf.Interval
Ctrl	4	1175000	262995.6	131497.8	22.38	756515.4, 1593485
Dose1	4	842500.0	241436.7	120718.3	28.66	458320.4, 1226680
Dose2	4	1112500	371068.3	185534.1	33.35	522047.6, 1702952
Dose3	4	1050000	288675.1	144337.6	27.49	590653.4, 1509347

Level	Median	Min	Max	%of Control(means)	%Reduction(means)
Ctrl	1250000	800000.0	1400000	.	.
Dose1	875000.0	520000.0	1100000	71.70	28.30
Dose2	1030000	790000.0	1600000	94.68	5.32
Dose3	1050000	700000.0	1400000	89.36	10.64

```
**************************************************************************
PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
    Analysis of Variance (ANOVA) - overall F-test
     Numerator df    Denominator df    F-stat       P-value
           3              12              0.96        0.445

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing negative trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level     Mean    Dunnett  Isotonic  Williams                 Tukey p-values
                  p-value    mean     p-value    Dose1   Dose2   Dose3   Dose4   Dose5

 Ctrl  1175000      .       1175000       .         .       .       .       .       .
 Dose1842500.0    0.302     1001667     0.252       .       .       .       .       .
 Dose2 1112500    0.981     1001667     0.270     0.584     .       .       .       .
 Dose3 1050000    0.879     1001667     0.279     0.755   0.990     .       .       .

**************************************************************************
NON-PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
    Kruskal-Wallis test - equality among treatment groups
     Degrees of Freedom   TestStat    P-value
           3                2.43       0.488

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing negative trend

 Level     Median          MannWhit p-value          Jonckheere p-value
  Ctrl    1250000                   .                        .
  Dose1  875000.0                 0.235                    0.074
  Dose2   1030000                 0.780                    0.304
  Dose3   1050000                 0.580                    0.372

 DECREASING TREND TEST SUMMARY      LOWEST CONCENTRATION SIGNIF. LESS THAN CONTROL
   Williams                                >highest dose (no sign. differences)
   Jonckheere                              >highest dose (no sign. differences)

**************************************************************************
PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
    Analysis of Variance (ANOVA) - overall F-test
     Numerator df    Denominator df    F-stat       P-value
           3              12              0.96        0.445

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing INCREASING trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level     Mean    Dunnett  Isotonic  Williams                 Tukey p-values
                  p-value    mean     p-value    Dose1   Dose2   Dose3   Dose4   Dose5

 Ctrl -1175000      .      -1008750      .         .       .       .       .       .
 Dose1 -842500    0.302    -1008750    0.855       .       .       .       .       .
```

Dose2-1112500	0.981	-1081250	0.789	0.584	.	.	.	.
Dose3-1050000	0.879	-1081250	0.807	0.755	0.990	.	.	.

****

NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	2.43	0.488

MannWhit - testing each trt median signif. different from control

Jonckheere - test assumes dose-response relationship, testing INCREASING trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	-1250000	.	.
Dose1	-875000	0.235	0.926
Dose2	-1030000	0.780	0.696
Dose3	-1050000	0.580	0.628

INCREASING TREND TEST SUMMARY	LOWEST CONCENTRATION SIGNIF. GREATER THAN CONTROL
Williams	>highest dose (no sign. differences)
Jonckheere	>highest dose (no sign. differences)

test for fish screen study - Cypermethrin

ANALYSIS RESULTS FOR VARIABLE VAR06 (M vitellogenin (ng/mL))

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS

Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01

Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05

Use parametric analyses if neither test rejected, otherwise non-parametric analyses.

Shapiro-Wilks Test Stat	Shapiro-Wilks P-value	Levenes Test Stat	Levenes P-value	Conclusion
0.857	0.017	2.309	0.128	USE PARAMETRIC TESTS

****

BASIC SUMMARY STATISTICS

Level	N	Mean	StdDev	StdErr	Coef of Var	95% Conf.Interval
Ctrl	4	41.00	33.34	16.67	81.31	-12.05, 94.05
Dose1	4	53.00	72.35	36.17	136.50	-62.12, 168.12
Dose2	4	39.25	25.38	12.69	64.67	-1.14, 79.64
Dose3	4	35.00	29.13	14.57	83.23	-11.36, 81.36

Level	Median	Min	Max	%of Control(means)	%Reduction(means)
Ctrl	38.00	12.00	76.00	.	.
Dose1	19.50	12.00	161.00	129.27	-29.27
Dose2	36.50	13.00	71.00	95.73	4.27
Dose3	25.00	13.00	77.00	85.37	14.63

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	0.12	0.946

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing negative trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	41.00	.	47.00	.	.	.	.	.	.
Dose1	53.00	0.962	47.00	0.662	.	.	.	.	.
Dose2	39.25	1.000	39.25	0.593	0.970	.	.	.	.
Dose3	35.00	0.995	35.00	0.551	0.938	0.999	.	.	.

****
NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	0.49	0.921

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing negative trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	38.00	.	.
Dose1	19.50	0.887	0.384
Dose2	36.50	1.000	0.588
Dose3	25.00	0.780	0.664

DECREASING TREND TEST SUMMARY — LOWEST CONCENTRATION SIGNIF. LESS THAN CONTROL
Williams >highest dose (no sign. differences)
Jonckheere >highest dose (no sign. differences)

****
PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	0.12	0.946

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing INCREASING trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	-41.00	.	-41.00	.	.	.	.	.	.
Dose1	-53.00	0.962	-42.42	0.564	.	.	.	.	.
Dose2	-39.25	1.000	-42.42	0.598	0.970	.	.	.	.
Dose3	-35.00	0.995	-42.42	0.617	0.938	0.999	.	.	.

****
NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value

3		0.49	0.921

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing INCREASING trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	-38.00	.	.
Dose1	-19.50	0.887	0.616
Dose2	-36.50	1.000	0.412
Dose3	-25.00	0.780	0.336

INCREASING TREND TEST SUMMARY	LOWEST CONCENTRATION SIGNIF. GREATER THAN CONTROL
Williams	>highest dose (no sign. differences)
Jonckheere	>highest dose (no sign. differences)

test for fish screen study - Cypermethrin
ANALYSIS RESULTS FOR VARIABLE VAR07 (F GSI)

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS
Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01
Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05
Use parametric analyses if neither test rejected, otherwise non-parametric analyses.

Shapiro-Wilks Test Stat	Shapiro-Wilks P-value	Levenes Test Stat	Levenes P-value	Conclusion
0.910	0.117	1.388	0.294	USE PARAMETRIC TESTS

****

BASIC SUMMARY STATISTICS

Level	N	Mean	StdDev	StdErr	Coef of Var	95% Conf.Interval	
Ctrl	4	11.88	4.21	2.11	35.46	5.17,	18.58
Dose1	4	11.28	2.18	1.09	19.37	7.80,	14.75
Dose2	4	11.20	2.25	1.12	20.06	7.63,	14.77
Dose3	4	14.25	1.50	0.75	10.53	11.86,	16.64

Level	Median	Min	Max	%of Control(means)	%Reduction(means)
Ctrl	10.50	8.50	18.00	.	.
Dose1	11.00	9.10	14.00	94.95	5.05
Dose2	10.85	9.10	14.00	94.32	5.68
Dose3	14.00	13.00	16.00	120.00	-20.00

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	1.10	0.386

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing negative trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5

Ctrl	11.88	.	12.15	.	.	.	.	.	.
Dose1	11.28	0.979	12.15	0.642	.	.	.	.	.
Dose2	11.20	0.971	12.15	0.677	1.000	.	.	.	.
Dose3	14.25	0.492	12.15	0.696	0.445	0.425	.	.	.

****

NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	3.82	0.282

MannWhit - testing each trt median signif. different from control

Jonckheere - test assumes dose-response relationship, testing negative trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	10.50	.	.
Dose1	11.00	1.000	0.558
Dose2	10.85	1.000	0.500
Dose3	14.00	0.343	0.933

DECREASING TREND TEST SUMMARY	LOWEST CONCENTRATION SIGNIF. LESS THAN CONTROL
Williams	>highest dose (no sign. differences)
Jonckheere	>highest dose (no sign. differences)

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	1.10	0.386

Dunnett - testing each trt mean signif. different than control

Williams - test assumes dose-response relationship, testing INCREASING trend

Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	-11.88	.	-11.45	.	.	.	.	.	.
Dose1	-11.28	0.979	-11.45	0.673	.	.	.	.	.
Dose2	-11.20	0.971	-11.45	0.708	1.000	.	.	.	.
Dose3	-14.25	0.492	-14.25	0.158	0.445	0.425	.	.	.

****

NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	3.82	0.282

MannWhit - testing each trt median signif. different from control

Jonckheere - test assumes dose-response relationship, testing INCREASING trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	-10.50	.	.

Dose1	-11.00	1.000	0.442
Dose2	-10.85	1.000	0.500
Dose3	-14.00	0.343	0.067

INCREASING TREND TEST SUMMARY — LOWEST CONCENTRATION SIGNIF. GREATER THAN CONTROL
Williams >highest dose (no sign. differences)
Jonckheere >highest dose (no sign. differences)

test for fish screen study - Cypermethrin
ANALYSIS RESULTS FOR VARIABLE VAR08 (M GSI)

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS
Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01
Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05
Use parametric analyses if neither test rejected, otherwise non-parametric analyses.

Shapiro-Wilks Test Stat	Shapiro-Wilks P-value	Levenes Test Stat	Levenes P-value	Conclusion
0.976	0.923	2.555	0.104	USE PARAMETRIC TESTS

****

BASIC SUMMARY STATISTICS

Level	N	Mean	StdDev	StdErr	Coef of Var	95% Conf.Interval	
Ctrl	4	1.11	0.25	0.12	22.28	0.71,	1.50
Dose1	4	1.22	0.18	0.09	14.72	0.93,	1.51
Dose2	4	1.35	0.06	0.03	4.28	1.26,	1.44
Dose3	4	1.40	0.18	0.09	13.04	1.11,	1.69

Level	Median	Min	Max	%of Control(means)	%Reduction(means)
Ctrl	1.09	0.84	1.40	.	.
Dose1	1.25	0.98	1.40	110.41	-10.41
Dose2	1.35	1.30	1.40	122.17	-22.17
Dose3	1.40	1.20	1.60	126.70	-26.70

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	2.18	0.143

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing negative trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	1.11	.	1.27	.	.	.	.	.	.
Dose1	1.22	0.700	1.27	0.939	.	.	.	.	.
Dose2	1.35	0.182	1.27	0.955	0.740	.	.	.	.
Dose3	1.40	0.095	1.27	0.962	0.515	0.978	.	.	.

****

NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

```
    Kruskal-Wallis test - equality among treatment groups
     Degrees of Freedom   TestStat    P-value
           3                4.28       0.233

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing negative trend

 Level     Median          MannWhit p-value          Jonckheere p-value
  Ctrl       1.09                   .                        .
  Dose1      1.25                0.575                    0.769
  Dose2      1.35                0.222                    0.959
  Dose3      1.40                0.190                    0.982

 DECREASING TREND TEST SUMMARY      LOWEST CONCENTRATION SIGNIF. LESS THAN CONTROL
   Williams                                >highest dose (no sign. differences)
   Jonckheere                              >highest dose (no sign. differences)

*****************************************************************************
PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
    Analysis of Variance (ANOVA) - overall F-test
     Numerator df    Denominator df    F-stat       P-value
           3             12              2.18       0.143

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing INCREASING trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level     Mean    Dunnett  Isotonic  Williams                    Tukey p-values
                  p-value    mean     p-value    Dose1   Dose2   Dose3   Dose4   Dose5

 Ctrl     -1.11     .       -1.11       .         .       .       .       .       .
 Dose1    -1.22   0.700     -1.22     0.229       .       .       .       .       .
 Dose2    -1.35   0.182     -1.35     0.048     0.740     .       .       .       .
 Dose3    -1.40   0.095     -1.40     0.024     0.515   0.978     .       .       .

*****************************************************************************
NON-PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
    Kruskal-Wallis test - equality among treatment groups
     Degrees of Freedom   TestStat    P-value
           3                4.28       0.233

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing INCREASING trend

 Level     Median          MannWhit p-value          Jonckheere p-value
  Ctrl      -1.09                   .                        .
  Dose1     -1.25                0.575                    0.231
  Dose2     -1.35                0.222                    0.041
  Dose3     -1.40                0.190                    0.018

 INCREASING TREND TEST SUMMARY      LOWEST CONCENTRATION SIGNIF. GREATER THAN CONTROL
   Williams                                Dose2
   Jonckheere                              Dose2
```

test for fish screen study - Cypermethrin
ANALYSIS RESULTS FOR VARIABLE VAR09 (F tubercle score (median))

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS
Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01
Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05
Use parametric analyses if neither test rejected, otherwise non-parametric analyses.

Shapiro-Wilks Test Stat	Shapiro-Wilks P-value	Levenes Test Stat	Levenes P-value	Conclusion
.	.	.	.	NO DATA FOR TEST

****
BASIC SUMMARY STATISTICS

Level	N	Mean	StdDev	StdErr	Coef of Var	95% Conf.Interval
Ctrl	4	0.00	0.00	0.00	.	. , .
Dose1	4	0.00	0.00	0.00	.	. , .
Dose2	4	0.00	0.00	0.00	.	. , .
Dose3	4	0.00	0.00	0.00	.	. , .

Level	Median	Min	Max	%of Control(means)	%Reduction(means)
Ctrl	0.00	0.00	0.00	.	.
Dose1	0.00	0.00	0.00	.	.
Dose2	0.00	0.00	0.00	.	.
Dose3	0.00	0.00	0.00	.	.

****
PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
.	1	.	.

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing negative trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	0.00	.	.	.	.	.	.	.	.
Dose1	0.00	.	.	.	.	.	.	.	.
Dose2	0.00	.	.	.	.	.	.	.	.
Dose3	0.00	.	.	.	.	.	.	.	.

****
NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	0.00	1.000

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing negative trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	0.00	.	.
Dose1	0.00	1.000	.
Dose2	0.00	1.000	.
Dose3	0.00	1.000	.

DECREASING TREND TEST SUMMARY LOWEST CONCENTRATION SIGNIF. LESS THAN CONTROL
Williams Dose1
Jonckheere Dose1

****
PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
.	1	.	.

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing INCREASING trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	0.00	.	.	.	.	.	.	.	.
Dose1	0.00	.	.	.	.	.	.	.	.
Dose2	0.00	.	.	.	.	.	.	.	.
Dose3	0.00	.	.	.	.	.	.	.	.

****
NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests
Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	0.00	1.000

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing INCREASING trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	0.00	.	.
Dose1	0.00	1.000	.
Dose2	0.00	1.000	.
Dose3	0.00	1.000	.

INCREASING TREND TEST SUMMARY LOWEST CONCENTRATION SIGNIF. GREATER THAN CONTROL
Williams Dose1
Jonckheere Dose1

test for fish screen study - Cypermethrin
ANALYSIS RESULTS FOR VARIABLE VAR10 (M tubercle score (median))

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS

Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01
Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05
Use parametric analyses if neither test rejected, otherwise non-parametric analyses.

Shapiro-Wilks Test Stat	Shapiro-Wilks P-value	Levenes Test Stat	Levenes P-value	Conclusion
0.967	0.782	1.418	0.286	USE PARAMETRIC TESTS

****

BASIC SUMMARY STATISTICS

Level	N	Mean	StdDev	StdErr	Coef of Var	95% Conf.Interval
Ctrl	4	32.50	2.08	1.04	6.41	29.19, 35.81
Dose1	4	37.75	5.91	2.95	15.65	28.35, 47.15
Dose2	4	32.25	4.65	2.32	14.41	24.86, 39.64
Dose3	4	26.50	4.51	2.25	17.02	19.32, 33.68

Level	Median	Min	Max	%of Control(means)	%Reduction(means)
Ctrl	32.50	30.00	35.00	.	.
Dose1	37.00	32.00	45.00	116.15	-16.15
Dose2	33.00	26.00	37.00	99.23	0.77
Dose3	25.00	23.00	33.00	81.54	18.46

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	4.16	0.031

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing negative trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	32.50	.	35.13	.	.	.	.	.	.
Dose1	37.75	0.279	35.13	0.861	.	.	.	.	.
Dose2	32.25	1.000	32.25	0.584	0.353	.	.	.	.
Dose3	26.50	0.194	26.50	0.053	0.019	0.318	.	.	.

****

NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	6.34	0.096

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing negative trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	32.50	.	.
Dose1	37.00	0.283	0.904
Dose2	33.00	1.000	0.559
Dose3	25.00	0.190	0.055

```
 DECREASING TREND TEST SUMMARY      LOWEST CONCENTRATION SIGNIF. LESS THAN CONTROL
   Williams                                 >highest dose (no sign. differences)
   Jonckheere                               >highest dose (no sign. differences)

*****************************************************************************
PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
    Analysis of Variance (ANOVA) - overall F-test
     Numerator df    Denominator df    F-stat       P-value
           3              12              4.16        0.031

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing INCREASING trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level     Mean    Dunnett  Isotonic  Williams                 Tukey p-values
                  p-value    mean     p-value    Dose1   Dose2   Dose3   Dose4   Dose5

 Ctrl    -32.50     .       -32.25       .         .       .       .       .       .
 Dose1   -37.75   0.279     -32.25     0.616       .       .       .       .       .
 Dose2   -32.25   1.000     -32.25     0.651     0.353     .       .       .       .
 Dose3   -26.50   0.194     -32.25     0.670     0.019   0.318     .       .       .

*****************************************************************************
NON-PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
    Kruskal-Wallis test - equality among treatment groups
     Degrees of Freedom   TestStat    P-value
           3                 6.34       0.096

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing INCREASING trend

 Level     Median          MannWhit p-value          Jonckheere p-value
  Ctrl     -32.50                    .                        .
  Dose1    -37.00                  0.283                    0.096
  Dose2    -33.00                  1.000                    0.441
  Dose3    -25.00                  0.190                    0.945

 INCREASING TREND TEST SUMMARY      LOWEST CONCENTRATION SIGNIF. GREATER THAN CONTROL
   Williams                                 >highest dose (no sign. differences)
   Jonckheere                               >highest dose (no sign. differences)

test for fish screen study - Cypermethrin
ANALYSIS RESULTS FOR VARIABLE VAR11   ( fecundity )

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS
Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01
Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05
Use parametric analyses if neither test rejected, otherwise non-parametric analyses.
  Shapiro-Wilks  Shapiro-Wilks    Levenes    Levenes   Conclusion
    Test Stat      P-value       Test Stat   P-value
      0.953         0.538          1.887      0.186    USE PARAMETRIC TESTS
```

```
**************************************************************************
BASIC SUMMARY STATISTICS
 Level  N     Mean     StdDev      StdErr    Coef of Var    95% Conf.Interval
  Ctrl  4    14.25      2.99        1.49       20.95          9.50,    19.00
  Dose1 4    13.50      3.00        1.50       22.22          8.73,    18.27
  Dose2 4     8.28      4.73        2.37       57.20          0.74,    15.81
  Dose3 4     2.23      0.88        0.44       39.42          0.83,     3.62

 Level       Median      Min       Max    %of Control(means)   %Reduction(means)
  Ctrl        14.00    11.00      18.00          .                    .
  Dose1       13.00    11.00      17.00        94.74                5.26
  Dose2        7.10     3.90      15.00        58.07               41.93
  Dose3        2.20     1.30       3.20        15.61               84.39

**************************************************************************
PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
    Analysis of Variance (ANOVA) - overall F-test
     Numerator df     Denominator df     F-stat       P-value
           3              12              12.07        <.001

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing negative trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level     Mean    Dunnett  Isotonic  Williams                 Tukey p-values
                  p-value    mean     p-value    Dose1   Dose2   Dose3   Dose4   Dose5

 Ctrl    14.25      .        14.25       .         .       .       .       .       .
 Dose1   13.50    0.975      13.50     0.442       .       .       .       .       .
 Dose2    8.28    0.054       8.28     0.013     0.151     .       .       .       .
 Dose3    2.23    <.001       2.23     <.001     0.002   0.083     .       .       .

**************************************************************************
NON-PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
    Kruskal-Wallis test - equality among treatment groups
     Degrees of Freedom   TestStat    P-value
           3                10.65       0.014

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing negative trend

 Level      Median          MannWhit p-value          Jonckheere p-value
  Ctrl       14.00                 .                        .
  Dose1      13.00               0.775                    0.328
  Dose2       7.10               0.190                    0.032
  Dose3       2.20               0.067                    <.001

 DECREASING TREND TEST SUMMARY      LOWEST CONCENTRATION SIGNIF. LESS THAN CONTROL
   Williams                                 Dose2
   Jonckheere                               Dose2

**************************************************************************
PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
```

Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	12.07	<.001

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing INCREASING trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	-14.25	.	-9.56	.	.	.	.	.	.
Dose1	-13.50	0.975	-9.56	0.988	.	.	.	.	.
Dose2	-8.28	0.054	-9.56	0.992	0.151	.	.	.	.
Dose3	-2.23	<.001	-9.56	0.994	0.002	0.083	.	.	.

****

NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	10.65	0.014

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing INCREASING trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	-14.00	.	.
Dose1	-13.00	0.775	0.672
Dose2	-7.10	0.190	0.968
Dose3	-2.20	0.067	1.000

INCREASING TREND TEST SUMMARY	LOWEST CONCENTRATION SIGNIF. GREATER THAN CONTROL
Williams	>highest dose (no sign. differences)
Jonckheere	>highest dose (no sign. differences)

test for fish screen study - Cypermethrin
ANALYSIS RESULTS FOR VARIABLE VAR12 (fertility)

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS
Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01
Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05
Use parametric analyses if neither test rejected, otherwise non-parametric analyses.

Shapiro-Wilks Test Stat	Shapiro-Wilks P-value	Levenes Test Stat	Levenes P-value	Conclusion
0.880	0.038	3.102	0.067	USE PARAMETRIC TESTS

****

BASIC SUMMARY STATISTICS

Level	N	Mean	StdDev	StdErr	Coef of Var	95% Conf.Interval
Ctrl	4	96.50	4.36	2.18	4.52	89.56, 103.44
Dose1	4	97.75	0.50	0.25	0.51	96.95, 98.55
Dose2	4	96.50	1.73	0.87	1.79	93.74, 99.26
Dose3	4	92.25	6.65	3.33	7.21	81.67, 102.83

Level	Median	Min	Max	%of Control(means)	%Reduction(means)
Ctrl	98.50	90.00	99.00	.	.
Dose1	98.00	97.00	98.00	101.30	-1.30
Dose2	97.00	94.00	98.00	100.00	0.00
Dose3	94.00	83.00	98.00	95.60	4.40

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	1.39	0.292

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing negative trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	96.50	.	97.13	.	.	.	.	.	.
Dose1	97.75	0.947	97.13	0.672	.	.	.	.	.
Dose2	96.50	1.000	96.50	0.618	0.972	.	.	.	.
Dose3	92.25	0.358	92.25	0.108	0.275	0.481	.	.	.

****

NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	4.32	0.229

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing negative trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	98.50	.	.
Dose1	98.00	0.555	0.219
Dose2	97.00	0.407	0.053
Dose3	94.00	0.281	0.010

DECREASING TREND TEST SUMMARY — LOWEST CONCENTRATION SIGNIF. LESS THAN CONTROL

Williams	>highest dose (no sign. differences)
Jonckheere	Dose3

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	1.39	0.292

Dunnett - testing each trt mean signif. different than control
Williams - test assumes dose-response relationship, testing INCREASING trend
Tukey - two-sided tests, all possible comparisons, not used for NOEC or LOEC

Level	Mean	Dunnett p-value	Isotonic mean	Williams p-value	Tukey p-values Dose1	Dose2	Dose3	Dose4	Dose5
Ctrl	-96.50	.	-95.75	.	.	.	.	.	.
Dose1	-97.75	0.947	-95.75	0.688	.	.	.	.	.
Dose2	-96.50	1.000	-95.75	0.723	0.972	.	.	.	.
Dose3	-92.25	0.358	-95.75	0.742	0.275	0.481	.	.	.

****

NON-PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Kruskal-Wallis test - equality among treatment groups

Degrees of Freedom	TestStat	P-value
3	4.32	0.229

MannWhit - testing each trt median signif. different from control

Jonckheere - test assumes dose-response relationship, testing INCREASING trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	-98.50	.	.
Dose1	-98.00	0.555	0.781
Dose2	-97.00	0.407	0.947
Dose3	-94.00	0.281	0.990

INCREASING TREND TEST SUMMARY	LOWEST CONCENTRATION SIGNIF. GREATER THAN CONTROL
Williams	>highest dose (no sign. differences)
Jonckheere	>highest dose (no sign. differences)

test for fish screen study - Cypermethrin

ANALYSIS RESULTS FOR VARIABLE VAR13 (F testosterone (ng/mL))

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS

Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01

Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05

Use parametric analyses if neither test rejected, otherwise non-parametric analyses.

Shapiro-Wilks Test Stat	Shapiro-Wilks P-value	Levenes Test Stat	Levenes P-value	Conclusion
.	.	.	.	NO DATA FOR TEST

****

BASIC SUMMARY STATISTICS

Level	N	Mean	StdDev	StdErr	Coef of Var	95% Conf.Interval
Ctrl	0	.	.	.	.	. , .
Dose1	0	.	.	.	.	. , .
Dose2	0	.	.	.	.	. , .
Dose3	0	.	.	.	.	. , .

Level	Median	Min	Max	%of Control(means)	%Reduction(means)
Ctrl	.	.	.	.	.
Dose1	.	.	.	.	.
Dose2	.	.	.	.	.
Dose3	.	.	.	.	.

```
**************************************************************************
PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
    Analysis of Variance (ANOVA) - overall F-test
     Numerator df    Denominator df    F-stat       P-value
           3            12               1.39        0.292

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing negative trend

 Level     Median          MannWhit p-value          Jonckheere p-value
  Ctrl        .                    .                        .
  Dose1       .                    .                        .
  Dose2       .                    .                        .
  Dose3       .                    .                        .
   Jonckheere                              Dose1

**************************************************************************
PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
    Analysis of Variance (ANOVA) - overall F-test
     Numerator df    Denominator df    F-stat       P-value
           3            12               1.39        0.292

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing INCREASING trend

 Level     Median          MannWhit p-value          Jonckheere p-value
  Ctrl        .                    .                        .
  Dose1       .                    .                        .
  Dose2       .                    .                        .
  Dose3       .                    .                        .
   Jonckheere                              Dose1

test for fish screen study - Cypermethrin
ANALYSIS RESULTS FOR VARIABLE VAR14   ( M testosterone (ng/mL) )

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS
Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01
Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05
Use parametric analyses if neither test rejected, otherwise non-parametric analyses.
  Shapiro-Wilks  Shapiro-Wilks    Levenes    Levenes   Conclusion
    Test Stat       P-value      Test Stat   P-value
       .              .             .          .       NO DATA FOR TEST

**************************************************************************
BASIC SUMMARY STATISTICS
 Level  N     Mean     StdDev      StdErr    Coef of Var     95% Conf.Interval
  Ctrl  0       .         .          .           .              .  ,     .
  Dose1 0       .         .          .           .              .  ,     .
  Dose2 0       .         .          .           .              .  ,     .
  Dose3 0       .         .          .           .              .  ,     .

 Level        Median      Min        Max     %of Control(means)    %Reduction(means)
  Ctrl           .         .          .           .                      .
```

Dose1	.	.	.	.	.
Dose2	.	.	.	.	.
Dose3	.	.	.	.	.

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	1.39	0.292

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing negative trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	.	.	.
Dose1	.	.	.
Dose2	.	.	.
Dose3	.	.	.

Jonckheere Dose1

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	1.39	0.292

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing INCREASING trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	.	.	.
Dose1	.	.	.
Dose2	.	.	.
Dose3	.	.	.

Jonckheere Dose1

test for fish screen study - Cypermethrin
ANALYSIS RESULTS FOR VARIABLE VAR15 (F 17b-estradiol (ng/mL))

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS
Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01
Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05
Use parametric analyses if neither test rejected, otherwise non-parametric analyses.

Shapiro-Wilks Test Stat	Shapiro-Wilks P-value	Levenes Test Stat	Levenes P-value	Conclusion
.	.	.	.	NO DATA FOR TEST

****

BASIC SUMMARY STATISTICS

Level	N	Mean	StdDev	StdErr	Coef of Var	95% Conf.Interval
Ctrl	0	.	.	.	.	. , .
Dose1	0	.	.	.	.	. , .
Dose2	0	.	.	.	.	. , .

```
  Dose3  0      .        .          .          .              .  ,     .

 Level       Median      Min       Max     %of Control(means)    %Reduction(means)
  Ctrl          .         .         .          .                      .
  Dose1         .         .         .          .                      .
  Dose2         .         .         .          .                      .
  Dose3         .         .         .          .                      .

*************************************************************************
PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
    Analysis of Variance (ANOVA) - overall F-test
     Numerator df     Denominator df     F-stat        P-value
           3             12                1.39         0.292

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing negative trend

 Level      Median          MannWhit p-value           Jonckheere p-value
  Ctrl         .                    .                        .
  Dose1        .                    .                        .
  Dose2        .                    .                        .
  Dose3        .                    .                        .
   Jonckheere                                Dose1

*************************************************************************
PARAMETRIC ANALYSES     - use alpha-level=0.05 for all tests
    Analysis of Variance (ANOVA) - overall F-test
     Numerator df     Denominator df     F-stat        P-value
           3             12                1.39         0.292

MannWhit - testing each trt median signif. different from control
Jonckheere - test assumes dose-response relationship, testing INCREASING trend

 Level      Median          MannWhit p-value           Jonckheere p-value
  Ctrl         .                    .                        .
  Dose1        .                    .                        .
  Dose2        .                    .                        .
  Dose3        .                    .                        .
   Jonckheere                                Dose1

test for fish screen study - Cypermethrin
ANALYSIS RESULTS FOR VARIABLE VAR16   ( M 17b-estradiol (ng/mL) )

TESTS OF ASSUMPTIONS FOR PARAMETRIC ANALYSIS
Shapiro-Wilks test for Normality of Residuals -- alpha-level=0.01
Levenes test for homogeneity of variance(absolute residuals) -- alpha-level=0.05
Use parametric analyses if neither test rejected, otherwise non-parametric analyses.
  Shapiro-Wilks  Shapiro-Wilks    Levenes    Levenes   Conclusion
    Test Stat      P-value       Test Stat   P-value
       .             .              .          .       NO DATA FOR TEST

*************************************************************************
BASIC SUMMARY STATISTICS
```

Level	N	Mean	StdDev	StdErr	Coef of Var	95% Conf.Interval
Ctrl	0	.	.	.	.	. , .
Dose1	0	.	.	.	.	. , .
Dose2	0	.	.	.	.	. , .
Dose3	0	.	.	.	.	. , .

Level	Median	Min	Max	%of Control(means)	%Reduction(means)
Ctrl	.	.	.	.	.
Dose1	.	.	.	.	.
Dose2	.	.	.	.	.
Dose3	.	.	.	.	.

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	1.39	0.292

MannWhit - testing each trt median signif. different from control

Jonckheere - test assumes dose-response relationship, testing negative trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	.	.	.
Dose1	.	.	.
Dose2	.	.	.
Dose3	.	.	.

Jonckheere Dose1

****

PARAMETRIC ANALYSES - use alpha-level=0.05 for all tests

Analysis of Variance (ANOVA) - overall F-test

Numerator df	Denominator df	F-stat	P-value
3	12	1.39	0.292

MannWhit - testing each trt median signif. different from control

Jonckheere - test assumes dose-response relationship, testing INCREASING trend

Level	Median	MannWhit p-value	Jonckheere p-value
Ctrl	.	.	.
Dose1	.	.	.
Dose2	.	.	.
Dose3	.	.	.

Jonckheere Dose1